Research Article

Formulation of Fast-Release Gastroretentive Solid Dispersion of Glibenclamide with Gelucire 50/13

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Abstract

Purpose: Fast-release gastroretentive solid dispersions of glibenclamide using gelucire were prepared to achieve improved bioavailability.

Methods: Hot melt granulation technique was adopted to prepare solid dispersions (SDs) of glibenclamide in gelucire 50/13 and were compared with pure glibenclamide and physical mixtures of drug and gelucire using hot stage polarized microscopy, powder x-ray diffraction (PXRD), Fourier-transform infrared spectroscopy FTIR, buoyancy as well as by in vitro release and in vivo studies. Further aging studies were carried out for the samples.

Results: PXRD showed that glibenclamide was present in SD in an amorphous form while FTIR spectroscopy revealed the presence of hydrogen bonding in the SDs. In vitro buoyancy was found for 11 h and there was improvement in solubility and dissolution rate for all test formulations. Formulations were found to follow Zero order kinetic. During aging study, no decrease of in vitro drug dissolution was observed over 3-month period. Crystallinity in the SDs was observed following aging. A more pronounced lowering of blood glucose level in Wistar rats compared with the pure drug, suggests that the test formulations are superior.

Conclusion: This study demonstrates the high potential of hot melt technique for obtaining stable fast-release gastroretentive solid dispersions of poorly water soluble drug using polyglycolized glycerides as carriers.

Keywords: Glibenclamide, Gelucire, Solid dispersion, Gastro-retentive multi-particulates, Hot melt technique.

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INTRODUCTION

The gastro-residence time of orally administered dosage form is generally short due to rapid gastric emptying. Rapid gastrointestinal transit could result in incomplete drug release from orally administered dosage form above the absorption zone, leading to diminished efficacy [1]. In order to increase the bioavailability of such drugs, the residence time of the orally administered dosage form in the upper gastrointestinal tract needs to be prolonged. The approaches to prolong gastroresidence time of pharmaceutical dosage forms include bioadhesive, mucoadhesive and density control delivery system [2-4].

Recently, much attention has been focused on the use of fats and fatty acid as carriers in drug delivery systems [5-7]. Mahadik et al [8] demonstrated the use of amphiphilic lipid glyceryl monooleate for the design of floating matrix system. Gelucire is in the family of vehicles derived from mixtures of mono-, di- and tri-glycerides with PEG esters of fatty acids. These are available with a range of properties depending on their hydrophilic-lipophilic balance (HLB) value and melting point range (33 - 65 ºC). These are used in the preparation of fast release and sustained release formulations. Gelucire containing only PEG esters are generally used in the preparation of fast release formulation. Owing to their extreme hydrophilicity and low density, Gelucire 50/13 may be considered an appropriate carrier for designing a fast release floating drug delivery system [9].

Solubility enhancement of glibenclamide (GLB) was achieved by dispersing the drug in molten Gelucire 50/13 at various ratios (1:1, 1:2, 1:4 and 1:10)[9]. It was poured on aluminium foil, allowed to solidify in a covered Petri dish and then kept in a refrigerator for 3 h. The solid lump was passed through a fine mesh (150 µm) to obtain a fine powder formulation that was then placed in a calcium chloride desiccator for 48 h. The quantities of GLB, SDs and hydrophilic additives, namely, polyethylene glycol (PEG) 200, 400, 4000 and 6000 used are as shown in Table 1. Preparation of solid dispersions (SDs) and physical mixtures (PMs)

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EXPERIMENTAL

Materials

Glibenclamide was received as a gift from Ranbaxy, Gurgaon, India. Gelucire 50/13 (semi-synthetic polyglycolized glycerides) was provided by Gattefosse, St.Priest, Cedex, France. All other materials and reagents used were of analytical grade.

Preparation of solid dispersions (SDs) and physical mixtures (PMs)

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Table 1: Composition (mg) of glibenclamide formulations containing Gelucire 50/13 and PEGs

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>GI 5</th>
<th>G II 5</th>
<th>G III 5</th>
<th>G IV 5</th>
<th>G V 5</th>
<th>G VI 5</th>
<th>G VII 5</th>
<th>G VIII 5</th>
<th>G IX 5</th>
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<tr>
<td>GLB</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
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<td>PEG 200</td>
<td>-</td>
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<td>PEG 400</td>
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<td>-</td>
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<td>10</td>
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<tr>
<td>PEG 4000</td>
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<td>5</td>
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<td>Gelucire 50/13</td>
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<td>60</td>
<td>65</td>
<td>65</td>
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</tr>
</tbody>
</table>

Determination of drug content of SDs

SDs equivalent to 5 mg of GLB were weighed accurately and dissolved in a suitable quantity of methanol. After filtration through a 0.45 µm filter (Ashless-Whatman, England), drug content was determined at 229 nm by spectrophotometry (Shimadzu UV-1800, Japan) with suitable dilution. Absorbance determination was performed in triplicate.

Evaluation of saturation solubility

The saturation solubility of GLB, PMs and SDs was evaluated by adding known excess amount of GLB formulation or pure GLB to 10 ml of 0.1M HCl (pH 1.2), stirred at 20 rpm in a water bath (25 ± 0.3 °C) for 48 h, filtered, diluted suitably with 0.1M HCl (pH 1.2) and analyzed at 229 nm.

Hot stage polarized microscopy (HSPM)

About 2 mg of sample was placed on a glass slide and covered with cover slip. The slide was examined under optical microscope fitted with a hot stage, heated at a rate of 2 °C/min from room temperature to 50 °C where it was held for 30 min, and then cooled to 40 °C. It was held at this temperature for 30 min. Polarized light microscopy was applied for the detection of crystallinity [13].

Powder x-ray diffraction (PXRD)

X-ray diffraction studies on GLB, Gelucire 50/13, PMs and SDs were determined using a D8 Advance, Bruker AXS instrument with a nickel-filtered radiation. The samples were irradiated with monochromatized CuK (α) radiation (1.542 Å) and analyzed between 2° and 50° 2θ using a step scan mode (step width = 0.020° (2θ), counting time = 0.5 s/step). Diffraction peak (d) intensities and 2h values of the SDs patterns were compared to those of the pure materials in order to evaluate the physical form of GLB in the samples.

Fourier-transform infrared spectroscopy (FTIR)

FTIR spectra of the individual materials as well as SDs were obtained, after appropriate background subtraction, using FTIR-8400 spectrometer (Shimadzu, Japan). About 2 – 3 mg of the sample was triturated with dry potassium bromide, compressed into disc and scanned from 4000 – 400 cm⁻¹.

Evaluation of in-vitro buoyancy

In vitro buoyancy assessment was performed using a USP dissolution apparatus type II by placing the SD in 900 ml of 0.1M HCl (pH 1.2) at 37.0 ± 0.5 °C and then agitated with a paddle at 75 rpm for 12 h. After agitation, the lipid particles that floated on the surface of the medium as well as those that settled at the bottom of the flask were removed separately. The proportion of floating particles was evaluated [14].

In vitro drug release studies

USP type II (paddle) method was used with the aid of Electrolab dissolution tester (TDT06 N, India). The dissolution medium
used was 900 mL of 0.1M HCl (pH 1.2) at 37 ± 0.5 °C and stirred at 75 rpm. An aliquot (5 ml) was withdrawn at predetermined time intervals for 4 h and replaced with same volume of fresh medium. The withdrawn sample was suitably diluted and analyzed using a spectrophotometer at 229 nm; the test was also carried out on a commercial brand of fast-release glibenclamide tablet (Betanase®, Cadila Healthcare Ltd, India).

**Analysis of release kinetic and drug release mechanism**

The release data obtained were treated according to zero-order, first-order, Higuchi [15] and Korsmeyer-Peppas’s models [16] with the aid of PCP-Disso software (V3, Poona College of Pharmacy, Pune, India), in order to analyze the kinetics of drug release from the formulations. Higuchi (Eq 1) and Korsmeyer-Peppas (Eq 2) models were also applied to determine drug release mechanism.

\[ Q = Kt^{1/2} \]  
where \( Q \) is the amount of drug released in time, \( t \), and \( K \) is the release constant.

\[ \frac{M_t}{M_\infty} = Kt^n \]  
where \( M_t/M_\infty \) is the fraction of drug released in time, \( t \), \( K \) is the structural and geometric constant, and \( n \) is the release exponent [17].

**Effect of ageing**

The SDs were stored at 30 °C/65 % RH for 3 months and the effect of ageing on the SDs were studied by measuring their in vitro release as well as structural features using DSC and XRPD.

**Evaluation of blood glucose**

Blood glucose level (BGL) lowering studies of the SDs and pure GLBs were determined in streptozotocin (STZ)-induced diabetic Wistar rats of either sex weighing 150 – 200 g. The animals were handled as per CPCSEA Guidelines of Good Laboratory Practice (GLP) [18]. They were housed in polypropylene cages with free access to standard laboratory diet and water. The research protocol of the animal experimentation (registration no. 837/ac/04/ CPCSEA, resolution no. 20/PhD/2008-2009, dated 8 March 2010) was approved by the ‘Institutional Animal Ethical Committee’ of College of Pharmacy, IFTM, Moradabad-244001, Uttar Pradesh, India. The animals were divided into 4 groups. Group I (control) was given 2 ml saline p.o. Diabetic control (Group II) was given STZ 35 mg/kg i.p. in citrate buffer (pH 4.5). Diabetic standard treatment Group III was given pure GLB (0.25 mg/kg) in aqueous solution p.o through an oral cannula. Diabetic test treatment (Group IV) was given GLB formulation (optimized batch) equivalent to 0.25 mg/kg of GLB p.o. in 2 ml of 1 % sodium carboxymethyl cellulose via an oral cannula. Blood glucose level (BGL) was measured periodically using Ascensia Entrust Glucometer (Bayer HealthCare, USA).

**Statistical analysis**

Statistical analysis of the data was carried out by Student’s t-test and one-way analysis of variance (ANOVA) at a significance level of \( p < 0.05 \) using SPSS 12.0 software (IBM).

**RESULTS**

**Saturation solubility**

The saturation solubility of GLB was 18.9 µg/ml while the enhanced saturated solubility obtained using drug:Gelucire (1:1) in physical mixture and solid dispersion was 27.23 and 44.39 µg/ml, respectively. Drug solubility increased in direct proportion to the proportion of Gelucire 50/13 in the preparations. Based on saturation solubility, ratio 1:10 showed enhanced solubility.

**Hot stage polarized microscopy (HSPM)**

HSPM of the SDs showed continuous melting from room temperature to 50 °C and when it was cooled back to 40 °C. Change in physical form was observed at different
Fig 1: Typical photomicrographs of solid dispersions (SDs) showing physical changes as temperature is varied.

Temperatures as shown in the photomicrographs in Fig 1. The photomicrographs indicate that large crystals of pure GLB were reduced to small particle size when they came in close contact with the hydrophilic carrier.

X-ray diffraction

The x-ray diffractograms of pure GLB (Fig 2a) had prominent diffraction peaks (d) equal to 8.035°, 7.464°, 4.649°, 3.860°, 2.933° and 1.687°, respectively on 2θ scale, which indicates its crystalline nature. The diffractograms for the formulations and Gelucire 50/13 (Fig 2b) indicate that GLB peaks decreased, suggesting its conversion from a crystalline to an amorphous state; diffractograms for the formulations physical mixtures and solid dispersions (Fig 2 (c) and (d)) indicate that GLB peaks decreased, suggesting its conversion from crystalline to an amorphous state; Gelucire 50/13 showed two prominent diffraction peaks (d) of 4.61° and 3.81° with the highest intensity on 2θ scale. The principal peaks of Gelucire 50/13
were present in both PMs and SDs with a lower intensity. The diffractogram of SDs showed absence of any trace of crystallinity, indicating the existence of amorphous GLB.

Fig 2: X-ray diffractogram of (a) pure GLB, (b) Gelucire 50/13, (c) physical mixtures (PMs) and (d) solid dispersions (SDs)

Fourier transform infrared (FTIR) spectra

FTIR spectra of pure GLB showed characteristic amide peaks at 3367.48, 3315.41, 2929.67, 2854.45 and 1716.53 cm\(^{-1}\); urea carbonyl stretching (urea NH stretching) vibration at 1618.2 and 1521.73 cm\(^{-1}\); and SO\(_2\) stretching vibration at 1340.43 and 1159.14 cm\(^{-1}\). On the other hand, the FTIR spectra of PMs showed slight intense amide peaks at 3315.41 and 1716.53 cm\(^{-1}\), while SDs showed almost complete disappearance of amide peaks at 3315.41 and 1716.53 cm\(^{-1}\), and concomitant shift to higher frequencies of urea carbonyl stretching vibration at 1618.2 and 1521.73 to 1635.5 and 1558.38 cm\(^{-1}\), respectively.

Fig 3: Comparative in vitro drug release of (A) SD formulations (◊ = G1, □ = G2, ○ = G3, △ = G4, □ = G5, ● = G6, × = G7, ▲ = G8; and (B) optimized SD (▲) and a commercial GLB brand, Betanase® (○).
Table 2: Release kinetic data for GLB solid dispersions (SDs) based on various models

<table>
<thead>
<tr>
<th>Formulation code</th>
<th>First order</th>
<th>Higuchi model</th>
<th>Zero order</th>
<th>Korsmeyer-Peppas</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>r²</td>
<td>n</td>
</tr>
<tr>
<td>GI</td>
<td>0.6893</td>
<td>0.9531</td>
<td>0.9874</td>
<td>0.9116</td>
</tr>
<tr>
<td>G II</td>
<td>0.6673</td>
<td>0.9152</td>
<td>0.9707</td>
<td>0.8939</td>
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<tr>
<td>G III</td>
<td>0.7200</td>
<td>0.9463</td>
<td>0.9722</td>
<td>0.9266</td>
</tr>
<tr>
<td>G IV</td>
<td>0.8005</td>
<td>0.9265</td>
<td>0.9888</td>
<td>0.9714</td>
</tr>
<tr>
<td>G V</td>
<td>0.7238</td>
<td>0.9750</td>
<td>0.9611</td>
<td>0.9334</td>
</tr>
<tr>
<td>G VI</td>
<td>0.7506</td>
<td>0.9210</td>
<td>0.9710</td>
<td>0.9462</td>
</tr>
<tr>
<td>G VII</td>
<td>0.6927</td>
<td>0.9732</td>
<td>0.9742</td>
<td>0.9131</td>
</tr>
<tr>
<td>G VIII</td>
<td>0.6959</td>
<td>0.9585</td>
<td>0.9915</td>
<td>0.9162</td>
</tr>
<tr>
<td>G IX</td>
<td>0.7072</td>
<td>0.8990</td>
<td>0.9288</td>
<td>0.9135</td>
</tr>
</tbody>
</table>

Note: Formulation code as in Table 1; r² = regression coefficient.

In vitro buoyancy, and drug entrapment and release

In vitro release profiles of the GLB SDs formulation in simulated gastric (pH 1.2) are shown in Fig 3A. In vitro buoyancy results indicate that the SD formulation remained floating for 11 h while drug entrapment efficiency was as high as 99.8%. The regression coefficient (r²) data based on kinetic analysis using various release models are listed in Table 2. The formulations were best fitted to the zero-order release model, with r² close to one. Korsmeyer-Peppas n data also indicate that the drug release mechanism was non-Fickian case II diffusion-controlled with n values ranging from 0.80 to 0.9221. The in vitro release data of optimized test GLB solid dispersion and commercial reference brand (Betanase® tablet) are indicated in Fig 3B. Release profiles of the two preparations were comparable.

Effect of ageing

When the GLB SDs were kept at 30 °C / 65 %RH for 3 months, no change in in vitro drug dissolution was observed, compared with the initial release rate.

Effect of GLB SDs on blood glucose level

As shown in Fig 4 the hypoglycemia produced in diabetic treated group IV treated with SD was significantly higher (p < 0.01) than the diabetic standard (GLB) treated group III. The Blood glucose level in diabetic treated group IV in 4 h were the same with Control group I compared with diabetic group II and diabetic standard treated group III.

![Blood Glucose Lowering Properties](image)

Fig 4: In vivo blood glucose lowering properties of optimized SD formulation in wistar rats. Key: (1) = Normal control, (2) = Diabetic control II, (3) = Diabetic standard treatment, and (4) Diabetic SD treatment
DISCUSSION

Hot melt granulation technique was selected to achieve solid dispersion, as it has been successfully utilized to increase the solubility of GLB [1]. PEGs and Gelucire are among the several carriers that have been employed in preparing solid dispersions.

The purpose of the current study was to examine the solid-state properties of a solid dispersion system of GLB prepared using Gelucire 50/13 and various grades of PEGs at varying ratios. Intermolecular interaction such as hydrogen bonding between the amide of GLB and the oxygen of polyglycol chain (Gelucire) inducing a shift of N-H vibration to an extent that depends on the strength of interaction. The site of the interaction on Gelucire would probably have been C=O group, which would also affect N-H vibration. This observation agrees with the data generated from PXRD and FTIR studies.

Both HSPM and x-ray diffraction analysis revealed that GLB was in an amorphous state and uniformly distributed throughout matrix while FTIR spectroscopy revealed the possibility of H-bonding interaction in both PMs and SDs.

To achieve gastro-retention, the time needed for the initiation of floatation (floating lag time) was less than 3 min for all the SD formulations. However, maximum in vitro buoyancy was 11 h. SDs exhibited dramatically improvement in initial rate as well as extent of in vitro drug dissolution. Zero-order kinetics model fitted best for the formulations while the n exponent of the Korsmeyer-Peppas model indicate that erosion was the mechanism of drug release. Release of drug from hydrophilic mini-matrices generally involves both pore diffusion and matrix erosion [5]. Low density and hydrophilic property due to the higher HLB of Gelucire 50/13 facilitated gastroretension and drug dissolution. Gelucire 50/13 possesses surfactant and self-emulsifying property and is also used as meltable binder by melt granulation of poorly water soluble active substances [9]. In contact with aqueous fluids, it forms a fine emulsion, solubilizes the active substance and hence increases its oral bioavailability.

Incorporation of GLB in the Gelucire 50/13 led to the formation of a solid dispersion system with increased dissolution rate of the drug due to improved wettability and the additional presence of a self-emulsifying compound, PEG. Moreover, it also increased the dispersability of the hydrophobic drug in the hydrophilic carrier during the process of solid dispersion formation. Thus, the increase in the dissolution of the drug can be attributable to improvement in wetting and to local solubilization by the excipients in the diffusion layer. The SDs exhibited higher burst release due probably to enhanced wettability of the drug particles, the emulsifying effect of carriers, significant reduction in particle size during SD formation and/or the inherently higher dissolution rate of soluble component of the SDs, which would pull along the more insoluble but finely mixed drug into the dissolution medium. Improved drug dissolution could also be attributed to the presence of the amorphous form of GLB, as indicated by the x-ray diffraction findings.

The increase in the release rate of the GLB SDs was also reflected in vivo by the greater reduction in blood glucose level in Wistar rats by GLB SDs, compared to pure GLB.

CONCLUSION

The present study demonstrates the high potential of hot melt-granulation technique for the production of solid dispersions of glibenclamide using polyglycolized glycerides as carriers. However, further studies are required to develop the formulation to industrial scale production.

COMPETING INTEREST

The authors declare no conflict of interest.
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